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What is claimed is:

1. A soluble proteic fragment of a subtilisin-kexin isoenzyme named SKI-1 which has the amino acid sequence defined by amino acids 187 to 996 of any one of SEQ ID NOs. 2, 4 and 6, and a variant thereof, which is enzymatically active.
- 5 2. A proteic fragment of a subtilisin-kexin isoenzyme named SKI-1, which has the amino acid sequence defined by amino acids 18 to 137 of any one of SEQ ID NOs: 2, 4 and 6, and a variant thereof, which is capable of binding with amino acids 18 to 1052 of SKI-1 in whole or in part.
- 10 3. The proteic fragment of claim 2, wherein said part has a molecular weight of about 14 KDa and forms a tight complex with the soluble fragment of SKI-1 as defined in claim 1.
- 15 4. The proteic fragment of claim 2, which is an inhibitor of SKI-1 activity.
5. The proteic fragment of claim 4, wherein the SKI-1 amino acid sequence that is modified to prevent further enzymatic processing in a cell expressing said proteic fragment.
- 16 6. The proteic fragment of claim 5, which is modified by amino acid substitution, deletion or rearrangement.
7. An isolated nucleic acid encoding a protein fragment as defined in claim 1.
8. An isolated nucleic acid encoding a proteic fragment as defined in claim 2.
- 20 9. An isolated nucleic acid encoding a proteic fragment as defined in claim 3.
10. An isolated nucleic acid encoding a proteic fragment as defined in any one of claims 4 to 6.
11. A recombinant vector comprising the nucleic acid defined in any one of claims 7 to 10.
- 25 12. The recombinant vector of claim 11, which is an expression vector.

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13. The recombinant vector of claim 12, which comprises a promoter expressible in a target cell wherein expression of said nucleic acid is desirable.

14. The recombinant vector of claim 12, which comprises an inducible promoter.

15. A recombinant host cell comprising the recombinant vector defined in any one of 5 claims 11 to 14.

16. A method of producing a proteic fragment of SKI-1 enzyme, which comprises the steps of:

culturing a recombinant host cell expressing a nucleic acid as defined in any one of claims 7 to 10 in a cell growth and expression-supportive culture medium; and 10 recovering said proteic fragment of SKI-1 in the culture medium.

17. A method for cleaving a substrate for SKI-1 enzyme, which comprises the step of:

a) contacting said substrate with a SKI-1 enzyme which has 1) an amino acid sequence defined by amino acids 18 to 1052 of any one of SEQ ID Nos: 2, 4, 6 and 15 an active variant thereof, or 2) a SKI-1 soluble fragment as defined in claim 1, or 3) a catalytic part of a) or b), or 4) a complex as defined in claim 3, for a time sufficient and in conditions adequate for such cleavage to occur,

with the proviso that said substrate is not a sterol-regulatory element-binding protein (SREBP).

18. A method for producing a protein or a peptide from a proteic precursor which is an 20 enzymatic substrate for SKI-1 enzyme, which comprises the steps of:

a) contacting said proteic precursor with a SKI-1 enzyme which has 1) an amino acid sequence defined by amino acids 18 to 1052 of any one of SEQ ID Nos: 2, 4, 6 and an active variant thereof, or 2) a SKI-1 soluble fragment as defined in claim 1, or 3) a catalytic part of a) or b), or 4) a complex as defined in claim 3, for a time sufficient 25 and in conditions adequate for such cleavage to occur; and

b) recovering said protein or peptide;

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with the proviso that said substrate is not a sterol-regulatory element-binding protein (SREBP).

19. The method of claim 17, which takes place in a cell or in the presence of a cellular population and wherein step a) comprises the step of transfecting a cell with a nucleic acid 5 expressing said SKI-1 enzyme.

20. The method of claim 19, wherein said cell expresses said proteic precursor or is transfected with a nucleic acid expressing said proteic precursor.

21. A method of inhibiting the activity of a subtilisin-kexin isoenzyme named SKI-1, which comprises the step of contacting SKI-1 with the inhibitor defined in any one of claims 10 4 to 6, 8 and 10.

22. A peptide of at least 7 amino acids capable of binding to and of being cleaved by SKI-1 catalytic site, comprising the following general formula:



wherein $\text{Xaa}_1, 2, 3$ and Z are any amino acid

15 J is an alkyl or aromatic hydrophobic amino acid

n is 1, 2 or 3

O is an acidic amino acid,

with the proviso that the peptide does not comprise the sequence Lys - Arg - Phe - Val - Phe - Asn - Lys - Ile - Glu.

20 23. A peptide as defined in claim 22, wherein Xaa_2 is Lys, Leu, Phe or Thr.

24. A peptide as defined in claim 23 which has the sequence:



25. A peptide as defined in any one of claims 22 to 24 which is labelled d.

26. A peptide as defined in claim 25 which is fluorogenic.

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27. A peptide as defined in claim 26 which is

Abz-Val-Phe-Arg-Ser-Leu-Lys-Tyr-Ala-Glu-Ser-Asp-Tyr(NO₂).

wherein

Abz is orthoaminobenzoic acid, and

5 Tyr(NO₂) is 3-nitrotyrosine.

28. The use of a peptide as defined in any one of claims 22 to 27 for monitoring the activity of a subtilisin-kexin isoenzyme named SKI-1.

29. The use as defined in any one of claims 22 to 27 for screening inhibitors of a subtilisin-kexin isoenzyme named SKI-1.

10 30. The use as defined in any one of claims 22 to 27 for screening a subtilisin-kexin isoenzyme named SKI-1.

31. The use of a peptide of at least 7 amino acids capable of binding to and of being cleaved by SKI-1 catalytic site, comprising the following general formula:

Arg Xaa₁ J Xaa₂ ↓ Xaa₃ (Z)_n O

15 wherein Xaa₁, 2, 3 and Z are any amino acid

J is an alkyl or aromatic hydrophobic amino acid

n is 1, 2 or 3

O is an acidic amino acid

for monitoring the activity of a subtilisin-kexin isoenzyme named SKI-1.

20 32. The use of a peptide of at least 7 amino acids capable of binding to and of being cleaved by SKI-1 catalytic site, comprising the following general formula:

Arg Xaa₁ J Xaa₂ ↓ Xaa₃ (Z)_n O

wherein Xaa₁, 2, 3 and Z are any amino acid

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J is an alkyl or aromatic hydrophobic amino acid

n is 1, 2 or 3

O is an acidic amino acid

for screening inhibitors or substrates of a subtilisin-kexin isoenzyme named SKI-1.

5 33. The use of an inhibitor of the activity of a subtilisin-kexin isoenzyme named SKI-1 in the making of a medication for treating a disease involving an overexpression of a SKI-1 or a SKI-1 substrate.

10 34. The use as defined in claim 33, wherein said disease is associated with any one of hypercholesterolemia, high levels of fatty acids, lipids or farnesyl pyrophosphate, liver steatosis, Ras-dependent cancer, restenosis and amyloid protein formation.

15 35. The use as defined in claim 33 or 34, wherein said inhibitor is defined in any one of claims 2, 4 to 6, 8 and 10.

20 36. A composition comprising a SKI-1 fragment as defined in any one of claims 1 to 6, or a nucleic acid defined in any one of claims 7 to 10, or a recombinant vector as defined in any one of claims 7 to 10, or a recombinant vector as defined in any one of claims 11 to 14.

37. The use of a SKI-1 enzyme as encoded by nucleic acids to 18 to 1052 of SEQ ID NOs: 1, 3 or 5, or of a catalytic part that is unique to SKI-1 enzyme, or of an active variant thereof, the nucleic acid of the variant sharing at least 70% homology with the nucleic acid defined in SEQ ID NOs.: 1, 3 and 5 and hybridizing therewith under stringent hybridization conditions, for cleaving a proteic precursor, with the proviso that said proteic precursor is not a sterol-regulatory element-binding protein (SREBP).